Journal of Research on the Lepidoptera 20(3): 170-173, 1981(83)

On the Supernumerary Chromosomes of *Tarache tropica* Guen. (Lepidoptera: Noctuidae)

P. K. Mohanty

and

B. Nayak

Department of Zoology, F. M. College, Balasore 756 001, Orissa, INDIA

Abstract. The karyotype of *Tarache tropica* Guen. consists of 1 to 7 minute dot-shaped supernumerary chromosomes in addition to the normal haploid complement, n=31. The exact nature of the accessory chromosomes, whether they are true supernumeraries or are the unpaired nonhomologous elements produced through interspecific hybridization, is not clear, though the latter explanation appears to be more probable.

Introduction

Many species of plants and animals show one or more supernumerary chromosomes, characteristically different from ordinary chromosomes in their karyotype (Wilson, 1925; White, 1973; Jones, 1975). In Lepidoptera, however, reports on the number of species showing such elements are rather scanty (Dederer, 1928; Beliajeff, 1930; Federley, 1938; Lorkovic, 1941; Maeki, 1953; de Lesse, 1960a; Bigger, 1976; Rao and Murty, 1976; Nayak, 1978). Indeed, it is difficult to distinguish genuine B-chromosomes since the lepidopteran karyotype often includes a large number of small dot-like compact chromosomes which yield very little information about their exact morphology. An attempt has been made here to analyze the nature of such supernumerary elements in the karyotype of *Tarache tropica*, a species cytologically investigated for the first time.

Material and Methods

The larvae of *Tarache tropica* were collected from their host plant (unidentified) in the close vicinity of Bhubaneswar during July-August, 1976. These were reared in cages in the laboratory. The mature male larvae (5th instar) and early pupae provided suitable testes material for the study of spermatocytic chromosomes. Temporary preparation of the material using Bellings' acetocaramine was tried. For permanent preparation, the larval sexual organs were dissected in a colchicine-hypotonic solution (0.01% Colchicine in 0.45% of Sodium Citrate solution), followed by washing in fresh hypotonic solution for 30-40 minutes at room temperature. The testis was then transferred to a slide which was slightly

tipped to remove the hypotonic solution completely. The material was then torn into pieces in a drop of 60% aceto-ethanol (1:3) fixative and then squashed. The slide was next placed in a freezing chamber of a freezer for 2-3 minutes and the cover glass removed with care using a razor blade. The frozen preparation was then subjected to gently blowing air over it. The slide was then transferred to absolute glacial acetic acid for ½ to 1 minute to remove cytoplasmic staining and the slide was again air dried. The preparation was stained with Giemsa, diluted about 50 times in Sorensens' phosphate buffer (pH 7) for 8-10 minutes at room temperature.

Observations

The diploid chromsomes, as revealed in some of the spermatogonial metaphase stages of *T. tropica*, were 2n=62. The individual chromosomes were dot-like, almost isodiametric and compact bodies yielding no information regarding the position of the centromere. Morphologically, the gonial chromosomes were not differentiable into sex-chromosomes and autosomes. The haploid chromosome number as established from a count of pro-metaphase or metaphase I chromosomes was n=31. Metaphase I bivalents appeared oval in a polar view and dumbbell-shaped in a equatorial view. Late diakinesis or pro-metaphase plates and metaphase I cells of different specimens when compared showed variation in chromosome numbers from specimen to specimen and from follicle to follicle of the same individual. In addition to the normal karyotype of n=31 bivalents, a variable number of minute chromosomal elements, much smaller than the normal bivalents, was encountered. The uneven distribution, irregular number, and minute size all indicated that they were not homologous to any of the members of the karyotype. They appeared to be univalents without any homologous partner. In 6 of the 21 individuals investigated, only one supernumerary element was observed in the karyotype. In five of these, the supernumerary chromosome was stable and was observed in every meiocyte, while in the sixth, some (possibly of one cyst) had single m-chromosome each which was not seen in the remainder. Metaphase II plates were rare. The m-chromosome frequency in other specimens was high, 3 to 7 elements were encountered at metaphase I. They were smaller than the regular chromosomes and showed irregular segregation at first anaphase. The exact nature of these minute elements, whether they are supernumeraries or are unpaired elements due to interspecific hybridization, is not clear. However, their uneven distribution indicates their supernumerary nature.

Discussion

In the absence of direct evidence, the origin of supernumeraries is highly enigmatic. White (1973) assumes their origin to be through fragmentation of heterochromatic blocks of normal chromosomes. Robinson (1971), as

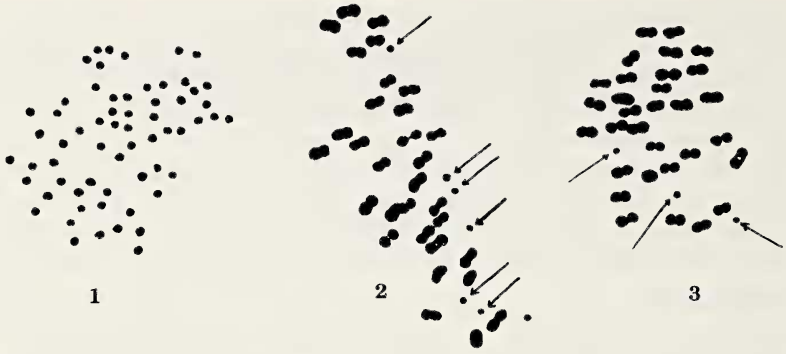

Fig. 1. Spermatogonial mitosis showing 62 dot-like elements.
Fig. 2. Metaphase I showing 31 bivalents and 7 extra elements of supernumerary nature.
Fig. 3. Metaphase I with 3 m-chromosomes.

well, holds them to be small segments of normal chromosomes undergoing accidental breakage and showing erratic meiotic behaviour due to their acentric nature. In Lepidoptera, where fragmentation is a common aspect of chromosome evolution, Robinson speculates that the occurrence of supernumeraries is no doubt a special case where an element is produced which behaves differently from a chromosome which has fragmented into two stable bodies, each of which behaves sufficiently normal to become a part of the karyotype. Jones (1975) is of the opinion that sex-chromosomes, particularly the X-chromosomes, are very likely the ancestors of micro-chromosomes. Bigger (1976) observed the karyotype of two British *Pieris* species to be identical in cells with and without B-chromosomes. Based on this observation, he concludes that supernumeraries are true additional chromosomes and not small fragments of the normal karyotype, as hypothesized by White (1973) and Robinson (1971). We agree with the last two authors that numerical variation may arise due either to accidental breakage of normal chromosomes or mixing of chromosomes from different nuclei. In the present case of *Tarache tropica*, the occurrence of 1 to 7 supernumeraries may well be due to interspecific hybridization in nature and may therefore present non-homologous unpaired univalents. Rao and Murty (1976) hypothesize that B-chromosomes confer an advantage on the population. This may not be true at least in the case of *Tarache tropica*, since a variable number of such supernumeraries occur in different follicles of the same individual, as well as in follicles of different individuals at the same time.

Acknowledgments. We thank Professor G. C. Das, Department of Zoology, Berhampur University, for valuable suggestions and one of us (P.K.M.) thanks V.G.C., New Delhi, for the financial assistance.

Literature Cited

BELIAJEFF, N. K., 1930. Die Chromosomenkomplexe und ihre Bezeihung zur Phylogenie bei Lepidopteren. Z. Ind. Abst. Vererbgsl. 54:369-399.

BIGGER, T. R. L., 1976. Karyotypes of three species of Lepidoptera including an investigation of B-chromosomes in *Pieris*. Cytologia 41:261-282.

DEDERER, P. H., 1928. Variation in the chromosome number in the spermatogenesis of *Philosamia cynthia*. Jour. Morph. 45:599-613.

FEDERLEY, H., 1938. Chromosomenzahlen finnlandischer Lepidopteren. I. Rhopalocera. Hereditas 24:221-269.

JONES, R. N., 1975. B-chromosome systems in flowering plants and animal species. Int. Rev. Cytol. Vol. 40:1-100.

LESSE, H. de, 1960a. Speciation et variation chromosomique chez les Lepidopteres Rhopaloceres. Ann. Sci. Nat. Zool. XII. Ser. 2:1-223.

LORKOVIC, Z., 1941. Die Chromosomenzahlen in der Spermatogenese der Tagfalter. Chromosoma 2:155-191.

MAEKI, K., 1953. A chromosome study of Japanese butterflies. Kwansei Gakuin Univ., Annual Stud. 1:67-70.

NAYAK, B., 1978. Supernumerary m-chromosomes in two species of Lepidoptera. Journ. of Entomology Research, Vol. 2, No. 1:112-114.

RAO, N. N. & A. S. MURTY, 1976. B-chromosomes in *Danaus limniace* (Lepidoptera: Danaidae). Microbios Letters, Vol. 3, No. 10.

ROBINSON, R., 1971. Lepidoptera Genetics, Pregamon Press, Oxford.

WHITE, M. J. D., 1973. Animal Cytology and Evolution, Cambridge University Press, London.

WILSON, E. B., 1925. The Cell in Development and Heredity, 3rd Ed. New York: MacMillan.